REMARKS

With entry of the amendment, claims 51-58 and 61 are pending; claims 59 and 60 are cancelled without prejudice or disclaimer. The cancellation of claims is not intended to be a dedication to the public of the subject matter canceled. Applicants reserve the right to file a continuation application directed to the canceled subject matter.

Claim 51 was amended to clarify that fluorescence of the fluorophore is reduced by fluorescence resonance energy transfer (FRET) to the quencher or by ground state quenching when the labeled oligonucleotide is not hybridized to the target nucleic acid, and that an increase in fluorescence is detected indicating the presence of the target nucleic acid sequence in the sample. Support for the amendment can be found at least at paragraphs [0003], [0051], and [0052]. Claims 56 and 58 were amended to replace fluorescent energy transfer with the art recognized term fluorescence resonance energy transfer, FRET, and claims 56-58 were amended to clarify that fluorescence is reduced when the oligonucleotide is not hybridized to the target nucleic acid sequence. Support for the amendment can be found at least at paragraphs [0003], [0051], and [0052]. Claim 61 was amended to correct its dependency, in light of the cancellation of claim 60, from which claim 61 formerly depended. Support for the amendment can be found in claims 51, 60, and 61. Thus, the amendments are fully supported by the specification and introduce no new matter.

Acknowledgement

Applicants wish to thank Examiner Staples, Primary Examiner Strzeleck, and Supervisor Benzion for their participation in an October 21, 2009 telephonic interview with Dr. Joe Walder, Attorney John Petravich, and the undersigned in which the invention and its patentability over the prior art was discussed.

Rejection of claims 51-60 under 35 USC §102

Claims 51-60 are rejected under 35 USC 102(b) as being anticipated by Batz (US Patent No. 6,117,973). The Examiner concluded that Batz teaches methods of detecting a target DNA in a sample using an oligonucleotide labeled with an acridine fluorophore, the fluorescence of which "can be reduced by energy transfer to the quencher or by ground state quenching by the quencher which is an anthraquinone moiety (see Example 11, especially column 36 lines 17-

49... wherein the quencher moiety can be an α-aminoanthraquinone according to formula VIa (see claim 7)".

Batz, as evidenced throughout the entire patent, is focused on compositions and methods for detecting a nucleic acid by binding a probe labeled with an electron donor, an electron acceptor, or both. The presence of the nucleic acid is detected by determining the transfer of an electron or hole from the electron donor to the electron acceptor (column 4, lines 11-28). Batz emphasizes that the invention overcomes disadvantages of FRET-based methods, which require overlap of the emission and absorption spectra of the fluorophor and quencher (column 3, lines 12-21). In this regard, Batz distinguishes electron transfer-based methods from the energy transfer-based methods of Tyagi (Nature Biotechnology 14:303-306, 1996), which describes the use of the quencher 4-(4'-dimethylaminophenylazo) benzoic acid (DABSYL), which is not an anthraquinone.

Example 11 of Batz describes using electron transfer quenching to detect hybridization (column 35, lines 63-64). Here Batz discloses that "the choice of electron transfer rather than energy transfer in the present invention arises from the fact that fewer restrictions are placed on the donor and acceptor moieties for photoinduced electron transfer chemistry. In particular either the donor or the acceptor can be irradiated and there is no requirement that the donor absorb light of shorter wavelength than the acceptor." (column 36, lines 10-16). Additionally, at column 21, lines 55-59, Batz states that "The ideal fluorescer will have ... energetics so that energy transfer to the anthraquinone is impossible but that electron transfer is fast." (Emphasis added). With respect to fluorescence quenching, energy transfer and electron transfer are competing processes.

Applicants' invention, in contrast to the disclosure of Batz, is directed to methods that involve quenching of the fluorescence of a fluorophore not by electron transfer, but by fluorescence resonance energy transfer or ground state quenching by an α-aminoanthraquinone. Applicants have discovered that by substituting an amino group at the alpha position of the anthraquinone nucleus the absorption spectra is shifted far to the red (see Figure 1) allowing the anthraquinone to efficiently quench the fluorescence of a broad range of fluorophores through either fluorescence resonance energy transfer or ground state quenching.

In Example 11, wherein Batz discloses the quenching of a probe labeled with the fluorophore acridine by electron transfer, the anthraquinone serves as an oxidizing agent, that is,

an electron acceptor. At column 11, lines 30-34, Batz teaches that for such electron acceptors substituents that are electron withdrawing are preferred. At column 15, line 63 to column 16, line 3, Batz lists substituents that are electron withdrawing and ones that are electron donating. Amino groups are specifically included in the list of electron donating substituents. Thus for the purpose of fluorescence quenching, Batz actually teaches away from the incorporation of amino groups within an anthraquinone.

In Example 11, "an anthraquinone moiety (Q1), which functions as an electron acceptor", was used (column 36, lines 20-21). Q1 was synthesized as described in Example 1; its structure is shown in Fig. 8. Clearly, Q1 is not an aminoanthraquinone, and the substituent attached to the anthraquinone nucleus is not at the alpha position but at the beta position. The substituent attached to the anthraquinone is a carboxyamide group which is electron withdrawing and is in fact included in the list of electron withdrawing substituents given by Batz. Clearly Batz provides no motivation to introduce electron donating groups, in general, and α-amino groups in particular within anthraquinones to enhance their use in fluorescence quenching.

The Examiner relies on claim 7 as purportedly disclosing α-aminoanthraquinone quenchers. Claim 7 depends from, and includes the limitations of claim 1, which encompasses a molecule of formula VIIIa, VIIIb, or VIIIc, each of which requires a moiety designated "L". Claim 1 defines "L" as being a non-nucleobase electron donor or acceptor moiety which is capable of participating in the complete transfer of an electron. (Column 45, lines 31-33).

Claim 7 requires that L have one of three general formulae that include VIa, a subgeneric structure encompassing millions of compounds. Nothing in the specification would lead one to at once envisage an α-aminoanthraquinone for the purpose of fluorescence quenching. Indeed Batz indicates just the opposite, that inclusion of an electron withdrawing substituent would be advantageous to enhance quenching. Furthermore, even if one had at once envisaged an α-aminoanthraquinone, claim 7 of Batz requires that L be capable of participating in the complete transfer of an electron. As noted above, quenching of fluorescence by fluorescence resonance energy transfer and fluorescence quenching by electron transfer are competing processes. As described in the present invention incorporation of an α-amino group within the anthraquinone greatly enhances fluorescence energy transfer, which by necessity, interferes with its participation in electron transfer. The requirement that "L" be capable of participating in the complete transfer of an electron, when taken in light of the emphasis on electron transfer

quenching by Batz, is in stark contrast to the Examiner's conclusion that claim 7 teaches α-aminoanthraquinone quenchers that inherently quench by energy transfer or ground state quenching.

It is important to point out that claim 7 of Batz, which is a composition claim, lists both electron withdrawing and electron donating substituents. Inclusion of both types of substituents stems from the fact that Batz describes a number of different assay formats some of which rely on fluorescence quenching by the anthraquinone and some dependent on other mechanisms (see for example column 19, lines 8-19). While it may be true that for certain of these alternative mechanisms the incorporation of an electron donating group would be beneficial, Batz clearly teaches that for the purpose of fluorescence quenching this would be undesirable and that an electron withdrawing substituent such as in Q1 would be preferred. The fact that incorporation of an α-amino group, an electron donating substituent, within the anthraquinone leads to enhanced quenching of fluorescence was completely unanticipated by Batz and results from the fact that α-aminoanthraquinones are effective fluorescent resonance energy transfer and ground state quenchers.

As for the Examiner's theory that the structures encompassed by claim 7 are inherently capable of energy transfer or ground state quenching, Batz did not synthesize or test an α-aminoanthraquinone as a fluorescence quencher or suggest the utility of an α-aminoanthraquinone as a fluorescence quencher. Claim 7 lists 24 different types of substituents that might be attached to an anthraquinone ring system. The only direction provided as to which might be preferred is that for the purposes of fluorescence quenching by electron transfer an electron withdrawing group would be desirable. This teaches away from anthraquinones containing electron donating substituents such as α-aminoanthraquinones. Moreover, the only anthraquinone actually studied by Batz was a derivative in which an electron withdrawing substituent was attached at the beta position (Q1). Hence, fluorescence quenching by an α-aminoanthraquinone through fluorescence resonance energy transfer or ground state quenching as described in the present invention was not inherently disclosed.

In light of the foregoing, Applicants request that the rejection be withdrawn.

Rejection of claim 61 under 35 USC §103(a)

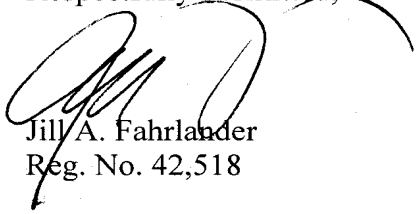
Claim 61 is rejected under 35 USC §103(a) as being unpatentable over Batz (US Patent No. 6,117,973), and Jenne (United States Patent No. 6,451,535). Batz is cited for the reasons

relied on for the rejection of claims under 35 USC §102(b). Jenne is cited as teaching a method for measuring RNase activity by detecting a change in fluorescence in a system comprising an RNA polymer labeled with a quencher and a fluorophore. Jenne does not cure the deficiencies of Batz in as much as it fails to teach the use of an oligonucleotide labeled with a fluorophore and an α-aminoanthraquinone quencher in which fluorescence of the fluorophore is reduced by fluorescence resonance energy transfer or by ground state quenching by an α-aminoanthraquinone.

Applicants believe that the present application is in condition for allowance. Favorable reconsideration of the application is respectfully requested. Should any questions remain, the Examiner is encouraged to contact the undersigned at **608.257.3501** so that prompt disposition of the application may be achieved.

Please charge the fee required for filing the Request for Continued Examination to Deposit Account No. **50-0842**. No other fee is believed due in connection with this submission. However, the Commissioner is authorized to charge any other fee which may be required to Deposit Account No. **50-0842**.

Respectfully submitted,



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